

originally-filed claims. Following entry of the present amendment, claims 49-60 are pending and presented for consideration.

The specification is hereby amended to include a specific reference to earlier filed applications for which the benefit of their filing dates is claimed under 35 U.S.C. §§119 and 120. Further amendments made to the specification correct typographical errors that one of skill in would recognize as such and would recognize the appropriate correction. Applicants submit that the present amendments introduce no new matter.

The Examiner is invited to call the undersigned if the Examiner believes that a telephone conversation could be helpful in expediting prosecution of the instant application.

Respectfully Submitted,



Maureen A. Bresnahan

Reg. No.: 44,559

Attorney for the Applicants

Testa, Hurwitz & Thibeault, LLP

High Street Tower

125 High Street

Boston, MA 02110

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Tel. No. (617) 248-7103

Fax No. (617) 248-7100

MARKED-UP VERSION OF REPLACEMENT PARAGRAPHS

~~{0001}~~ [0002] The present invention relates to a medicine, drink, food [and] or feed having an action of strengthening bone. The medicine, drink, food or feed which is combined with collagen, fraction containing collagen, and/or degradation product that has effects of promoting bone formation, strengthening bone, preventing and treating bone metabolic diseases such as osteoporosis, bone fracture, bone pain and so on.

[0002] [0003] Accompanying [with] the prolongation of the human life span, the incidence of metabolic bone diseases such as osteoporosis, bone fracture, bone pain etc., has recently increased. In bone tissue, bone formation and bone resorption are always occurring. While the balance of bone formation and bone resorption is kept in one's youth, bone resorption exceeds bone formation due to various causes as one's age increases (uncoupling). And when bone resorption [prolongs] persists for a long duration, bone tissue becomes fragile, which causes metabolic bone diseases such as osteoporosis, bone fracture, bone pain, etc. [, are expected to be prevented.]

~~{0003}~~ [0004] [As conventional] Conventional methods of preventing or treating metabolic bone diseases by inhibiting uncoupling[.] include (1) calcium supplemented diets, (2) light exercise, (3) sunbathing, (4) medicinal therapy, etc., are exemplified. As for calcium supplemented diets, calcium salts such as calcium carbonate, calcium phosphate, etc., and naturally occurring calcium-containing preparation, such as bovine bone powder, egg shell, fish bone powder, etc. are used. They are, however, not necessarily good enough for oral intake. [As light] Light exercises, jogging or walking may be recommended. However, they are troublesome to a person who becomes weak and quite difficult to an immobilized aged person. Sunbathing is believed to be good for [supplement of] supplementing the active form of vitamin D₃ but is not sufficient. As medicinal therapy, 1 α -hydroxyvitamin D₃ and/or calcitonin may be used and they are known to be effective for treating osteoporosis. However, [they are medicines themselves and] these

~~{0004}~~ [0005] On the other hand, collagen is known as a main protein component [composing] of animal connective tissue and occupies nearly 30% of total protein in mammalian, especially, human whole body. This collagen is a protein of cellular matrix and can be defined to be a substance having an α -chain which is a helical portion consisting of 3 polypeptide chains and forming a multi-molecular complex. Further, collagen forms intercellular matrix with glycoproteins, such as proteoglycan, fibronectin, laminin etc., and is an essential component for exhibiting its function as assembled tissue of cells in multi-cellular biological organism.

~~{0005}~~ [0006] The present inventors have investigated to find [out] a substance [having action] capable of strengthening bone which is useful for food sources. Eventually, we found out that a fraction containing collagen such as skin protein fraction or bone protein fraction has an effect of promoting proliferation of an osteoblast. Further, the present inventors found that degradation products of the fraction containing collagen also have an effect of promoting proliferation of an osteoblast and accomplished the present invention.

~~{0006}~~ [0007] Accordingly, an object of the present invention is to provide a medicine, drink, food [and] or feed having an action of strengthening bone. More specifically, the present invention is to provide a medicine, drink, food and feed having an action of strengthening bone combined with collagen, fraction containing collagen and/or degradation product thereof.

~~{0007}~~ [0008] Another object of the present invention is to provide a medicine, drink, food [and] or feed combined with collagen, fraction containing collagen or degradation product thereof which is further combined with calcium and vitamins.

~~{0007}~~ [0009] As a fraction containing collagen used in the present invention, skin protein

lyophilization product of pulverized and decalcified bone, and fraction which is produced by treating skin protein fraction or bone protein fraction with acid or [alkaline] alkaline can be exemplified. Further, a degradation produce which is produced by hydrolysis with a proteolytic enzyme and has a molecular weight of 2-150 kDa can be used.

~~{0011}~~ [0012] In the present invention, collagen, a fraction containing [fraction] collagen and/or degradation product thereof can be combined with a medicine, drink, food or feed for having an action of strengthening bone. This fraction containing collagen is a protein fraction obtained from skin or bone. For example, when it is prepared from skin, skin can be depilated and corium layer was cut off. The tissue can be cut into pieces with a high-speed cutter, defatted with organic solvent and lyophilized to give a fraction containing collagen. [And when] When it is prepared from bone, bone can be decalcified with acid etc., pulverized with a high-speed cutter, decalcified, again, with acid etc., and lyophilized to give a fraction containing collagen. Collagen can be [more] purified by treating skin protein fraction or bone protein fraction containing collagen with acid or alkaline. Treatment with acid or alkaline can be carried out by soaking these fractions in a liquid solution containing 5-30 % [of liquid solution] of acid, such as hydrochloric acid or nitric acid, or alkaline, such as sodium hydroxide or potassium hydroxide, at 20-50°C for 0.5-3 hours. After soaking, these fractions should be sufficiently washed with water. Hot water extraction of these fractions can be carried out by treating them with 70-80°C hot water.

~~{0013}~~ [0014] In addition, calcium or vitamins can be combined with these fractions containing collagen. As a calcium source, calcium chloride, calcium carbonate, calcium lactate, egg shell or milk-derived calcium can be used. As combination rate of calcium with fraction containing collagen, 0.5-5.0 weight parts of said fraction will be preferable to 1 weight part of calcium. As vitamins, any vitamin can be used [and] however vitamin D₃ [or] a substance containing vitamin D₃ [can be more] is preferably used. As raw material containing substantial amount of collagen, fresh skin or bone of cattle, pig, chicken, sheep and horse can be [exemplified] utilized

~~{0014}~~ [0015] In the present invention, collagen, fraction containing collagen and/or degradation product thereof can be combined with a medicine, such as tablet or powder, [and] or with a drink or food, such as milk, yogurt, ice cream, milk drink, coffee drink, juice, jelly, noodle cracker, bread or sausage, for endowing an action of strengthening bone. Further, in the same manner, these fractions or degradation produce can be combined with a feed for endowing an action of strengthening bone. In addition, calcium [agent] agents with good absorptivity, such as calcium chloride, calcium carbonate, calcium lactate, egg shell or milk-derived calcium, etc., or vitamin D can be combined so that the action of strengthening bone thereof will be enhanced.

~~{0015}~~ [0016] Since a medicine, drink, food or feed combined with collagen, fraction containing collagen and/or degradation product thereof has an action of strengthening bone, oral administration of these will be useful for prevention and/or [improvement] treatment of bone metabolic diseases such as osteoporosis etc.[.]

~~{0016}~~ [0017] Preparation of fractions containing collagen and degradation products thereof, and an action of strengthening bone will be described by exemplifying reference examples and test examples [as] set forth below. Further, the present invention will be illustrated by [described by exemplifying] examples. However, these examples will not limit the scope of the present invention.